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Dietary starch-lipid ratio modifies femoral bone marrow adiposity but not tibial micro-architecture and biomechanics in 35d-old meat-type chickens

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Abstract

If the current trend in cereal grain supply and pricing is sustained, fats may replace carbohydrates as the preferred calorie supply for poultry diets, raising concerns over the potential impact of high-fat diets on the skeletal health of poultry, particularly meat-type chickens. Against this backdrop, the effect of diets with different starch-lipid ratios on growth and bone quality of 35d meat-type chickens was investigated. One hundred and eighty 1-day old, unsexed broiler chicks were offered either a high fat (HF) or low fat (LF) diet with starch-lipid ratios of 3:1 and 10:1, respectively. The experimental diets were replicated five times, with 18 chicks per replicate, in a two- phase feeding regimen: starter, 0-21 days, and grower, 22-35 days. Growth of birds was monitored, and pectoralis major, tibia, and femur were obtained from two chickens per replicate at 35d post-hatch. The pectoralis major was assayed for crude fat and protein, and the tibia assayed for linear and geometric morphometrics, biomechanical indices, and bone ash. Histomorphometric assessment was conducted on tibia for microarchitectural indices; real bone volume (BV/TV), trabecular separation (Tb.Sp), trabecular thickness (Tb.Th), and number of trabeculae (Tb.N), and on femoral marrow to assess femoral marrow adiposity.

Starch-lipid ratios of diets had no significant influence on body weight, crude fat and protein content of pectoralis major, bone ash, tibial morphometrics, and biomechanical indices, and trabecular microarchitecture except for tibial cross-sectional area which was lower in chickens on HF diet (13.08 vs 14.7mm^2, $p = 0.04$). However, a higher adiposity number per femoral marrow area (157.02 vs. 135.49mm^2, $p < 0.001$) was recorded and higher osteoclast activity was visually observed on the trabecular surfaces in chickens on the HF diet, indicative of a higher occurrence of bone resorption. It is imperative to conduct more studies to ascertain the potential implications of the observed increase in osteoclast activity and bone marrow adiposity on the skeletal health of meat-type chickens during and after the growth phase.

Keywords: Dietary energy; meat-type chickens; high fat diets; bone quality; bone marrow adipose tissue

Short running title: Starch-lipid ratios and bone quality of meat-type chickens

Le rapport amidon-lipides alimentaires modifie l'adiposité de la moelle osseuse fémorale, mais pas la microarchitecture et la biomécanique tibiales chez des poulets à viande de 35 jours

Résumé

Si la tendance actuelle en matière d'approvisionnement et de prix des céréales se maintient, les graisses pourraient remplacer les glucides comme apport calorique préféré pour l'alimentation des volailles, ce qui soulève des inquiétudes quant à l'impact potentiel des régimes riches en graisses sur la santé du squelette des volailles, en particulier des poulets à viande. Dans ce contexte, l'effet de régimes avec différents rapports amidon-lipides sur la croissance et la qualité osseuse des poulets de type viande 35d a été étudié. Cent quatre-vingt poussins de chair non sexués âgés d'un jour ont reçu un régime riche en graisses (RG) ou faible en graisses (FG) avec des rapports amidon-lipides de 3: 1 et 10: 1, respectivement. Les régimes expérimentaux ont été répétés cinq fois, avec 18 poussins par répétition, dans un régime alimentaire en deux phases : démarrage, 0-21 jours, et croissance, 22-35 jours. La croissance des oiseaux a été surveillée et le grand pectoral, le tibia et le fémur ont été obtenus à partir de deux poulets par répétition à 35 jours après l'éclosion. Le grand pectoral a été analysé pour la graisse brute et les protéines, et le tibia a été analysé pour la morphométrie linéaire et géométrique, les indices biomécaniques et les cendres osseuses. Une évaluation histomorphométrique a été réalisée sur le tibia pour les indices microarchitecturaux ; volume osseux *réel (BV/TV), séparation trabéculaire (Tb.Sp), épaisseur trabéculaire (Tb.Th) et nombre de trabécules (Tb.N), et sur la moelle fémorale pour évaluer l'adiposité de la moelle fémorale. Les rapports amidon-lipides des régimes n'ont eu aucune influence significative sur le poids corporel, la teneur en matières grasses brutes et en protéines du grand pectoral, les cendres osseuses, la morphométrie tibiale et les indices biomécaniques, et la microarchitecture trabéculaire, à l'exception de la section transversale tibiale qui était plus faible chez les poulets sous HF. alimentation (13,08 contre 14,7 mm2, p = 0,04). Cependant, un nombre plus élevé d'adiposité par zone de moelle fémorale (157,02 contre 135,49 mm2, p < 0,001) a été enregistré et une activité ostéoclastique plus élevée a été visuellement observée sur les surfaces trabéculaires chez les poulets sous régime RG, ce qui indique une fréquence plus élevée de résorption osseuse. Il est impératif de mener davantage d'études pour déterminer les implications potentielles de l'augmentation observée de l'activité des ostéoclastes et de l'adiposité de la moelle osseuse sur la santé du squelette des poulets de type viande pendant et après la phase de croissance.*

Mots-clés : Énergie alimentaire ; poulets à viande; régimes riches en graisses; qualité osseuse; tissu adipeux de la moelle osseuse

Introduction

Starch and lipids are major calorie contributors in conventional broiler diets, with typical diets containing up to 50% starch (Svihus, 2014) mainly supplied by wheat or corn depending on geographical location (Adeleye and Oladotun., 2020). The supply of these cereal grains is threatened by droughts, floods, declining crop yields, and more recently supply chain disruptions. These result in supply shortages and higher prices of these cereal grains. While chickens are considered more efficient at utilizing starch for energy (Aderibigbe *et al*., 2020; Truong *et al*., 2015) as a result of their developed capacity for starch digestion *in ovo* and rapid secretion of amylase and disaccharidases post hatch (Iji *et al*., 2001), higher starch concentrations in diets are believed to raise carcass lipid concentrations (Khoddami *et al*., 2018) and a reduction in dietary starch levels is proposed to optimize starch digestion and deter enteric disorders stimulated by ileal microbial proliferation (Engberg *et al*. 2004; Yun, *et al*., 2000; Hauck 2017). Lipid-rich feedstuff such as tallow, lard, and soyabean oil are often incorporated in broiler formulations to replace wheat and maize when they are in short supply or beef up the caloric value of diets as lipids have 2.25 times stored energy per mole than starch (Allahyari-Bake and Jahanian, 2017; Itani *et al*., 2020; Leeson and Summers, 2005). Despite contrary evidence to depressed lipid digestion in chicks owing to low lipase activity and delayed onset of bile acid secretion, increased body weight (Malheiros *et al*. 2004) and carcass weight, with decreased diet cost (Tabeidian *et al*., 2010) have been reported in response to diets with lower starch-lipid ratios in meat-type chickens. While the ability of high fat diets to impair calcium absorption has been established in mice (Macri *et al*. 2012; Xiao *et al*. 2010) and chicken studies (Atteh *et al*., 1989; Atteh and Leeson, 1984), its influence on skeletal health in chickens is still scarcely reported (Jingying *et al*., 2021; Wei *et al*., 2021). It is well documented that a significant fraction of whole-body energy needs is utilized within the skeletal niche involving bone cells, skeletal stem cells and bone marrow adipose tissues (Al-Bari and Al Mamun, 2020; Rendina-Ruedy and Rosen, 2020; Riddle and Clemens, 2017), with studies involving humans and animal models of obesity and anorexia nervosa showing that the bone marrow adipose tissue is an endocrine-active fat depot (Piotrowska and Tarnowski, 2021) and responds to metabolic changes arising from nutritional therapies such as caloric restrictions (Devlin *et al*., 2010; Spurny *et al*., 2020), dietary fat types (Hassan *et al*., 2019) and concentration. Hence, this study tests the hypothesis that diets with a lower starch-lipid ratio may impair skeletal integrity in 35d old meat-type chickens using bone marrow adiposity, tibial micro-architecture, and tibial biomechanics as markers.

Materials and methods

The present study is one of a series of research in which the ability of dietary starch-lipid ratios to modify post-hatch growth performance and development in meat-type chickens was investigated. The study was conducted at the Poultry Unit, Teaching and Research Farm, University of Ibadan, Nigeria. All animal welfare and handling procedures were in strict compliance with ‘ethical guidelines for research in Animal Science’, and EU standards for protection of animals, and feed legislation (Jarvis *et al*., 2005; National Research Council (US) Committee for the Update of the Guide for the Care and Use of Laboratory Animals 2011). Protocols were reviewed and approved by Department of Animal Science, University of Ibadan (Ibadan, Nigeria), and reporting conformed to stipulations in “Animal Research: Reporting of *in vivo* experiments” (ARRIVE) guidelines 2.0 (https://arriveguidelines.org).

The feeding trial comprised of two isocaloric and isonitrogenous diets with varied starch-lipid ratios: 3:1 (High fat, HF) and 10:1 (Low fat, LF). The HF and LF diets consisted of 8.6% fat and 20.56% starch + sugar and 3.87% fat and 33.2% starch + sugar, respectively in starter phase, 9.69% fat and 29.37% starch + sugar and 4.3% fat and 39.29% starch + sugar, respectively in grower phase. Most lipid calories in HF diet were supplied from soyabean in form of full fat soya and soya oil. Diets were formulated to meet breeders' specifications for starter and grower meat-type chickens as shown in Tables 1 and 2.

A total of 180-day-old unsexed broiler chicks were obtained from a commercial hatchery and randomly assigned to one of two dietary treatments for a period of 35d. Treatments consisted of 5 replicates of 18 chicks per replicate and random allocation to the treatments was based on weight equalization using Experimental Animal Allotment Program (Kim and Lindemann, 2007). Chicks were raised on deep litter with wood shavings as bedding material. A room temperature of 32ºC was maintained for the first seven days and thereafter an ambient temperature of 24-29ºC until the end of the study. A lighting program of 24L: 0D was observed during the first seven days and thereafter a 16L: 8D until end of the study. Water and mash feed were offered ad libitum, with starter diets fed from 1-21d and grower diets fed from 22-35d.

Table 1. Gross composition and calculated nutrients of experimental starter diet for meat-type chickens

Ingredients (g/1000g)	Low fat (LF) diet	High fat (HF) diet
Starch-lipid ratio	10:1	3:1
Soya bean meal	315.00	168.80
Full fat soya	-	180.00
Spray dried plasma	20.00	20.00
Maize	480.00	257.50
Corn bran	137.00	302.20
Soya oil	10.00	33.00
Limestone	8.20	4.00
Dicalcium phosphate	17.00	17.5
Choline chloride	1.00	1.00
Sodium bicarbonate	1.00	1.00
Vitamin mineral premix	2.50	2.50
Lysine HCl	2.50	2.90
Dl-methionine	2.00	2.00
L- threonine	0.80	0.70
NaCl	2.00	2.00
Total	1000.00	1000.00
Phytase	+	+
Carbohydrase	+	+
Protease	+	+
Calculated analysis		
Metabolizable energy (kcal/kg)	2855	2850
Metabolizable energy contribution from lipids (kcal/kg (%))	340.96 (11.96%)	751.52 (26.37%)
Crude protein (%)	21.70	21.70
Crude fat (%)	3.50	7.88
Crude fibre (%)	4.26	4.96
Starch + sugar (%)	33.27	20.56
Calcium (%)	0.91	0.92
Available phosphorus (%)	0.48	0.46
Non-phytate phosphorus	0.46	0.46
Methionine (%)	0.51	0.5
Digestible methionine (%)	0.46	0.47
Lysine (%)	1.25	1.27
Digestible lysine (%)	1.12	1.14
Threonine (%)	0.69	0.78

Composition of vitamin mineral premix per kg/diet: vitamin A, 10,000 IU; vitamin D3, 2,000 IU; vitamin E, 32 mg; vitamin K3, 1.6 mg; vitamin B1, 2.5 mg; vitamin B2, 4.4 mg; niacin, 44 mg; calcium pantothenate, 9.2 mg; vitamin B6, 4 mg; vitamin B12, 0.02mg; choline chloride, 400 mg; folic acid, 0.8 mg; biotin, 0.064 mg; manganese, 96 mg; iron, 80 mg; zinc, 64 mg; copper, 6.8 mg; iodine, 1.2 mg; cobalt, 0.24 mg; selenium, 0.096 mg; antioxidant, 96 mg.

Ronozyme Multigrain DSM N.V, Netherlands, included at 120g/tonne of feed, Ronozyme Hiphos DSM N.V, Netherlands, included at 500g/ tonne of feed, Ronozyme proAct DSM N.V, Netherlands, included at 200g/ tonne of feed

Calculated analysis based on nutritional values of feed ingredients (NRC 1994; Feedipedia).

Table 2. Gross composition and calculated nutrients of experimental grower diet for meat-type chickens

Ingredient (g/1000g)	Low fat (LF) diet	High fat (HF) diet
Starch-lipid ratio	10:1	3:1
Soya bean meal	279.00	90.50
Full fat soya	-	233.50
Spray dried plasma	20.00	20.00
Maize	512.10	250.00
Corn bran	140.90	327.00
Soya oil	16.50	43.50
Limestone	4.80	3.00
Dicalcium phosphate	14.60	20.00
Choline chloride	1.00	1.00
Sodium bicarbonate	1.00	1.00
Vitamin mineral premix	2.50	2.80
Lysine HCl	3.00	2.90
Dl-methionine	1.80	2.10
L- threonine	0.80	0.70
NaCl	2.00	2.00
Total	1000	1000.00
Phytase	+	+
Carbohydrase	+	+
Protease	+	+
Calculated analysis		
Metabolizable energy (kcal/kg)	2945.00	2945.00
Metabolizable energy contribution from lipids (kcal/kg (%))	409.18 (13.90%)	923.93 (31.38%)
Crude protein (%)	20.43	20.43
Crude fat (%)	4.30	9.69
Crude fibre (%)	3.98	4.79
Starch + sugar (%)	39.29	29.37
Calcium (%)	0.84	0.82
Available phosphorus (%)	0.42	0.43
Non-phytate phosphorus	0.42	0.43
Methionine (%)	0.49	0.5
Digestible methionine (%)	0.44	0.45
Lysine (%)	1.20	1.21
Digestible lysine (%)	1.08	1.08
Threonine	0.69	0.78

Composition of vitamin mineral premix per kg/diet: vitamin A, 10,000 IU; vitamin D_3, 2,000 IU; vitamin E, 32 mg; vitamin K_3, 1.6 mg; vitamin B_1, 2.5 mg; vitamin B_2, 4.4 mg; niacin, 44 mg; calcium pantothenate, 9.2 mg; vitamin B_6, 4 mg; vitamin B_{12}, 0.02mg; choline chloride, 400 mg; folic acid, 0.8 mg; biotin, 0.064 mg; manganese, 96 mg; iron, 80 mg; zinc, 64 mg; copper, 6.8 mg; iodine, 1.2 mg; cobalt, 0.24 mg; selenium, 0.096 mg; antioxidant, 96 mg.

Ronozyme Multigrain DSM N.V, Netherlands, included at 120g/ tonne of feed, Ronozyme Hiphos DSM N.V, Netherlands, included at 500g/ tonne of feed, Ronozyme proAct DSM N.V, Netherlands, included at 200g/ tonne of feed

Calculated analysis based on nutritional values of feed ingredients (NRC 1994; Feedipedia).

On d35, two chickens per replicate were individually weighed, sacrificed, and exsanguinated. Left and right tibia and femur were dissected from each carcass and stripped of muscles and connective tissues. Right tibiae were assayed for morphometric traits including weight, length, Seedor index, as well as geometric and mechanical properties (Adeleye *et al*., 2021; Guo *et al*., 2019; Seedor *et al*., 1991).

Left tibiae were collected and preserved in 10% neutral-buffered formaldehyde until analyses commenced. Tibiae were decalcified, dehydrated, and embedded in wax, from which 5mm thick sagittal specimen sections were prepared. Specimen were stained with hematoxylin-eosin to visualize the trabecular microarchitecture.

Femoral bone marrow was isolated first by removing epiphysis by lateral incisions with a blade to allow access to bone marrow. Subsequently, diaphysis was bisected, and bone marrow was scrapped out using a stainless-steel spatula. Bone marrow tissue was fixed in 10% neutral-buffered formaldehyde, paraffin embedded, sectioned, and stained with hematoxylin-eosin (Parlee *et al*., 2014).

Mechanical properties of the right tibiae were evaluated, and thereafter weighed, oven dried, milled and reweighed. Milled bones were defatted and air dried, and 0.5g aliquots were weighed in duplicate and burned in a muffle furnace at 600°C (AOAC, 2000) to determine bone ash. Microscopic images of tibial growth plate and femoral marrow were captured with use of a software Olympus cellSens version 1.5 (Olympus, Tokyo, Japan). Tibia trabecular microstructure was assessed for real bone volume (BV/TV), trabecular separation (Tb.Sp), trabecular thickness (Tb.Th), and number of trabeculae (Tb.N), and femoral marrow adiposity quantified using BoneJ plugin of ImageJ software (Wayne Rasband, NIMH, Bethesda, MD, USA), and degree of trabecular bone resorption was visually assessed. All data were subjected to a student's t-test procedure of JASP statistical software (version 0.14.1) (JASP, 2020) and significance was based on a 5% probability level.

Results

Chickens on the HF diet exhibited similar body weight, as well as fat and protein composition of *pectoralis major* ($p > 0.05$) at 35d, as chickens on the control (LF) diet (Table 3).

Table 3. Growth, *pectoralis major* composition, plus physical, geometrical and mechanical properties of 35d old meat-type chickens fed diets with varied starch-lipid ratios

Dietary treatments	Low fat (LF) diet	High fat (HF) diet	SEM	Probability
Starch-lipid ratio	10:1	3:1		
Initial body weight at 1d, g	40.50	40.23	0.17	0.34
Final body weight at 35d, g	490.80 ±92.83	465.35 ±88.64	14.31	0.38
Fat composition of *pectoralis major*, %	0.12 ± 0.08	0.12 ± 0.11	0.03	0.97
Protein composition of *pectoralis major*, %	22.75 ± 1.24	22.52 ± 0.41	0.28	0.71
Bone ash (%)	46.00 ± 2.94	45.50 ± 1.92	0.82	0.79
Linear measurements				
Tibia weight (g/100g BW)	0.97 ± 0.19	1.01 ± 0.25	0.07	0.80
Tibia length (mm/100g BW)	13.71 ± 1.39	14.71 ± 1.27	0.42	0.37
Seedor index	71.85 ± 16.42	69.84 ± 16.54	4.93	0.85
Bone index (g/kg BW)	11.19 ± 2.05	9.07 ± 3.23	0.88	0.25
Geometric measurements				
Horizontal external diameter (mm)	5.45 ± 0.61	5.16 ± 0.54	0.18	0.49
Vertical external diameter (mm)	4.87 ± 0.80	4.56 ± 0.80	0.26	0.579
Horizontal internal diameter (mm)	3.31 ± 0.66	2.63 ± 0.88	0.28	0.24
Vertical internal diameter (mm)	2.40 ± 0.42	2.63 ± 0.45	0.14	0.46
Cross sectional area (mm^2)	14.70 ± 3.64	13.08 ± 2.48	0.99	0.04
Mean relative wall thickness (mm)	0.85 ± 0.16	0.91 ± 0.19	0.06	0.67
Cortical index	45.03 ± 4.62	46.05 ± 4.99	1.52	0.76
Moment of inertia (mm^4)	31.30 ± 16.97	24.13 ± 15.92	5.26	0.54
Index of gyration (mm)	1.40 ± 0.25	1.29 ± 0.27	0.09	0.55
Mechanical properties				
Tibial breaking strength (kN/s)	0.84 ± 0.05	0.81 ± 0.03	0.03	0.59

Linear and geometric measurements, and mechanical properties of tibia of 35d chickens on both diets did not differ significantly except for tibial cross-sectional area which was lower in chickens on HF diet compared to the control (13.08 vs 14.7mm^2, $p = 0.04$ Table 3). Histological assessments of left tibiae growth plate also showed similar BV/TV proportions (real bone volume), trabecular number (Tb.N), trabecular thickness (Tb.Th) and trabecular spacing (Th.Sp) in chickens on both HF and LF diets (Figure 1). However, visual assessment of heamatoxylin and eosin-stained sections of trabecular bone in proximal tibia (Figure 2) show more trabecular resorption in HF treatment owing to higher osteoclast activity in this group. Adiposity number per marrow area of femur of chickens fed experimental diets (Figure 3A and 3B) was also significantly influenced by diet types, as chickens on low fat (LF) diet showed fewer adipocytes per marrow area (135.49 vs. 157.02mm^2, $p < 0.001$).

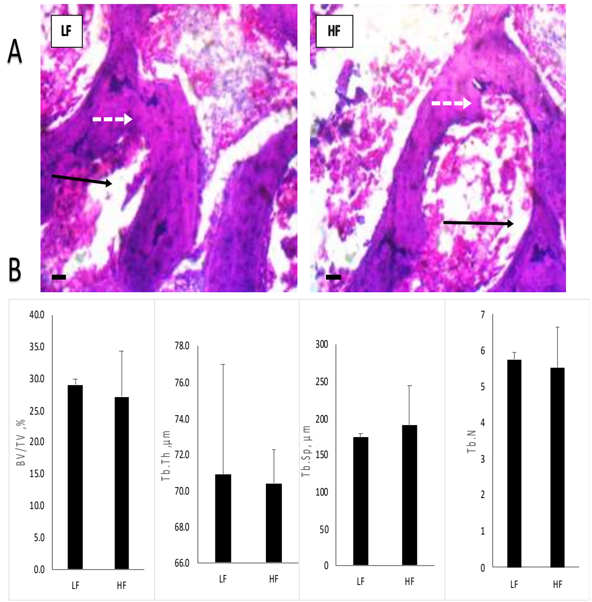

Figure 1. Characteristics of tibial head (growth plate) of 35d old meat-type chickens fed diets with varied starch-lipid ratios (a) representative images of heamatoxylin and eosin staining carried out on formaldehyde-fixed sections of trabecular bone. Red arrows indicate trabecular thickness (Tb.th), black arrows indicate trabecular spacing (Tb.Sp). (b) Real bone volume (BV/TV), trabecular thickness (Tb.Th), trabecular spacing (Tb.Sp), and number of trabeculae (Tb.N). LF – low fat (10:1) and HF – high fat (3:1) diets. All scale bars represent 50 µm.

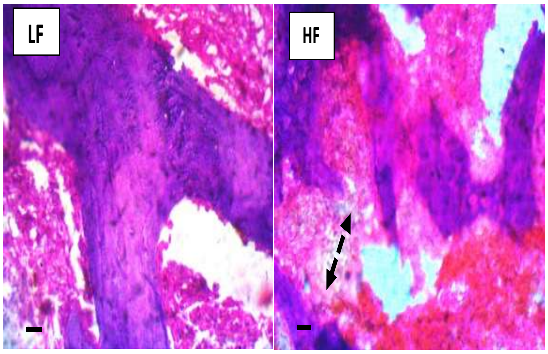

Figure 2. Histomorphometry of trabecular bone in the proximal tibia of 35d-old meat-type chickens fed diets with varied starch-lipid ratios. Representative images of heamatoxylin and eosin staining carried out on formaldehyde-fixed sections of trabecular

bone of the tibia. Black arrow indicates area of trabecular bone resorption. LF – low fat (10:1) and HF – high fat (3:1) diets. All scale bars represent 50 µm

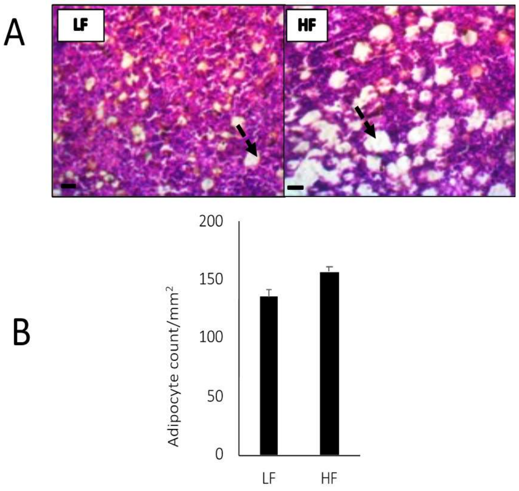

Figure 3. Histomorphometry of the femoral marrow of 35 days - old meat-type chickens fed diets with varied starch-lipid ratios. Representative images of A. heamatoxylin and eosin staining carried out on formaldehyde-fixed sections of the femoral bone marrow. Black arrows indicate adipocytes, and C. Mean adipocyte count in femoral bone marrow, LF – low fat (10:1) and HF – high fat (3:1) diets. All scale bars represent 50 µm.

Discussion

An increment in lipid to starch ratio of poultry diets is often aimed at increasing the energy density of feeds to support growth development in the chickens. While a diet is often adjudged "high fat" when >30% of its total energy constituents are supplied by lipids (Qiao *et al*., 2021), its implications for growth and skeletal development in meat-type chickens is still under-researched. In contrast to the current study, high fat diets are reported to improve body weight and feed efficiency, whilst reducing feed cost in meat-type flocks (Tabeidian *et al*., 2010; Atteh and Leeson, 1984; Wei *et al*., 2021), without modifying the lipid and protein constituents of broiler carcass (Khoddami *et al*., 2018). Yet, high fat diets are also reported to impair mineral metabolism due to formation of insoluble "soaps" by complexing of fatty acids and minerals such as magnesium, calcium, and zinc (Atteh *et al*.,1989). It is a shared opinion that modern meat-type chicken are more prone to disorders of bone growth and fractures due to stress emanating from a high body weight to bone ratio attained over a short time period which predisposes them to leg weakness and ultimately mortality stemming from starvation and dehydration (Bradshaw *et al*., 2002; Fleming, 2008; Santos *et al*., 2022; Thorp, 1994). However, according to Fleming (2008), these challenges are less evident when slower-growing strains are used. Findings of Wohl *et al*., (1998) and Harash *et al*., (2020) corroborate the current study showing no significant effects of high fat diets on the growth pattern, mechanical properties, geometric structure, and mineral content of bones of roosters and meat-type chickens, although Wohl *et al*., (1998) observed significant reduction in trabecular bone composition which could be attributed to reduction in bone calcium of meat-type chickens fed high fat diets (Atteh *et al*., 1984). More frequently, similar research conducted in mice models of human diseases such as obesity, diabetes and atherosclerosis show that dietary administration of diets with high lipid fractions compromise bone quality. The trabecular bone has been recognized to respond more to changes in diet types than the cortical bone due to its metabolic functions, with studies in mice showing that bone volume, trabeculae number, trabecular thickness and separation are negatively affected by increasing fat intake levels (Cao *et al*., 2020; Cao *et al*., 2009; Scheller *et al*., 2016). The absence of a response of trabecular indices to high fat diets in the current study could be attributed to length of the study period, and slow-growing *nature* of the study animals. However, the onset of bone resorption arising from increased osteoclast activity in chickens on the high fat diets observed in this study could be seen as a metabolic response induced by the high fat diet (Tencerova *et al*. 2018). Bone marrow adipose tissue development is a normal physiological process, and is considered an important biomarker of compromised skeletal integrity (Scheller *et al*., 2016). BMAT is quantified by static indices such as adipocyte number, adipocytes count and percentage adipocyte volume per tissue volume, and these indices have been shown to respond to sex, age and dietary manipulation in animals (Bukowska *et al*., 2018; Horowitz *et al*., 2017; Suchacki and Cawthorn, 2018). High fat diets induce the expansion of the bone marrow adipocytes in mice and human studies (Fonseca *et al*., 2021; Shu *et al*., 2015) and enhances osteoclastic activity resulting in bone resorption and decalcification of bone as observed in this study (Sutton *et al*., 2015; Theill *et al*., 2002).

This study's findings conclude that while feeding diets with a low starch-lipid ratio to meat-type chickens in the starter and grower stages did not adversely influence growth performance, linear and geometric measures, mechanical qualities, or trabecular microarchitecture, however, histomorphometry assessments showed visible trabecular bone resorption and an increase in the number of adipose cells per femoral marrow area. Possible implications of these for skeletal health during and beyond the growers' phase should be further investigated using more refined skeletal integrity indices.

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